



PCT/GB 2004 / 0 0 1 4 6 4  
Rec'd PCT/PTO 04 OCT 2005  
18/552895  
INVESTOR IN PEOPLE

The Patent Office  
Concept House  
Cardiff Road  
Newport  
South Wales  
NP10 8QQ

REC'D 04 JUN 2004

WIPO

PCT

## PRIORITY DOCUMENT

SUBMITTED OR TRANSMITTED IN  
COMPLIANCE WITH RULE 17.1(a) OR (b)

I, the undersigned, being an officer duly authorised in accordance with Section 74(1) and (4) of the Deregulation & Contracting Out Act 1994, to sign and issue certificates on behalf of the Comptroller-General, hereby certify that annexed hereto is a true copy of the documents as originally filed in connection with the patent application identified therein.

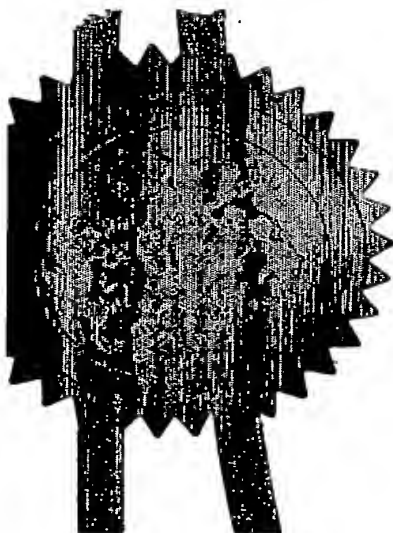
In accordance with the Patents (Companies Re-registration) Rules 1982, if a company named in this certificate and any accompanying documents has re-registered under the Companies Act 1980 with the same name as that with which it was registered immediately before re-registration save for the substitution as, or inclusion as, the last part of the name of the words "public limited company" or their equivalents in Welsh, references to the name of the company in this certificate and any accompanying documents shall be treated as references to the name with which it is so re-registered.

In accordance with the rules, the words "public limited company" may be replaced by p.l.c., plc, P.L.C. or PLC.

Re-registration under the Companies Act does not constitute a new legal entity but merely subjects the company to certain additional company law rules.

Signed *Andrew*

Dated 7 May 2004



BEST AVAILABLE COPY

**THE PATENT OFFICE**  
A  
12 AUG 2003  
NEWPORT



12AUG03 0829536-1 010099  
P01/7700 0-00-0318871.1

**Request for grant of a patent**

(See the notes on the back of this form. You can also get an explanatory leaflet from the Patent Office to help you fill in this form)

The Patent Office

Cardiff Road  
Newport  
South Wales  
NP10 8QQ

1. Your reference	10/722/P/GB		
2. Patent application number (The Patent Office will fill in this part)	12 AUG 2003	0318871.1	
3. Full name, address and postcode of the or of each applicant (underline all surnames)	Tissuemed Limited 5 Killingbeck Drive Leeds LS14 6UF		
Patents ADP number (if you know it)			
If the applicant is a corporate body, give the country/state of its incorporation	England	7499346002	
4. Title of the invention	Tissue-adhesive formulations		
5. Name of your agent (if you have one)	Adamson Jones		
"Address for service" in the United Kingdom to which all correspondence should be sent (including the postcode)	Broadway Business Centre 32a Stoney Street Nottingham NG1 1LL		
Patents ADP number (if you know it)	07979487001		
6. If you are declaring priority from one or more earlier patent applications, give the country and the date of filing of the or of each of these earlier applications and (if you know it) the or each application number	Country	Priority application number (if you know it)	Date of filing (day / month / year)
7. If this application is divided or otherwise derived from an earlier UK application, give the number and the filing date of the earlier application	Number of earlier application		Date of filing (day / month / year)
8. Is a statement of inventorship and of right to grant of a patent required in support of this request? (Answer 'Yes' if	Yes		
a) any applicant named in part 3 is not an inventor, or			
b) there is an inventor who is not named as an applicant, or			
c) any named applicant is a corporate body.			
See note (d))			

# Patents Form 1/77

9. Enter the number of sheets for any of the following items you are filing with this form. Do not count copies of the same document

Continuation sheets of this form

Description	
Claim(s)	28
Abstract	6
Drawing(s)	0
	3 + 3

10. If you are also filing any of the following, state how many against each item.

Priority documents

Translations of priority documents

Statement of inventorship and right to grant of a patent (Patents Form 7/77)

Request for preliminary examination and search (Patents Form 9/77)

Request for substantive examination (Patents Form 10/77)

Any other documents (please specify)

11. I/We request the grant of a patent on the basis of this application.

Signature

Date

11 August 2003

12. Name and daytime telephone number of person to contact in the United Kingdom

Dr S A Jones

0115 9247 147

## Warning

After an application for a patent has been filed, the Comptroller of the Patent Office will consider whether publication or communication of the invention should be prohibited or restricted under Section 22 of the Patents Act 1977. You will be informed if it is necessary to prohibit or restrict your invention in this way. Furthermore, if you live in the United Kingdom, Section 23 of the Patents Act 1977 stops you from applying for a patent abroad without first getting written permission from the Patent Office unless an application has been filed at least 6 weeks beforehand in the United Kingdom for a patent for the same invention and either no direction prohibiting publication or communication has been given, or any such direction has been revoked.

## Notes

- If you need help to fill in this form or you have any questions, please contact the Patent Office on 08459 500505.
- Write your answers in capital letters using black ink or you may type them.
- If there is not enough space for all the relevant details on any part of this form, please continue on a separate sheet of paper and write "see continuation sheet" in the relevant part(s). Any continuation sheet should be attached to this form.
- If you have answered 'Yes' Patents Form 7/77 will need to be filed.
- Once you have filled in the form you must remember to sign and date it.
- For details of the fee and ways to pay please contact the Patent Office.

## Title – Tissue-adhesive formulations

### Field of the Invention

5 This invention relates to materials suitable for use as tissue adhesives and sealants, and to a flexible multilamellar sheet, patch or film comprising such materials for topical application to internal and external surfaces of the body, for therapeutic purposes, to a process for the preparation of such products, and to methods of using such products. In particular the invention relates to  
10 materials that are formulated as loose or compacted powders and to a self-adhesive, biocompatible and hydratable polymeric sheet with such materials applied to a suitable support, which may be used for therapeutic purposes such as wound healing, joining, sealing and reinforcing weakened tissue, and for drug delivery, and to a process for preparing, and methods of using, such  
15 a sheet.

### Background of the Invention

There is considerable interest in the use, for a number surgical or other  
20 therapeutic applications, of materials that adhere to biological tissues eg as an alternative to the use of mechanical fasteners such as sutures, staples etc. Formulations of such materials that have hitherto been proposed include viscous solutions or gels that are either manufactured in that form or are prepared immediately prior to use by mixing of the ingredients. Such  
25 formulations are then applied to the tissue surface using a suitable applicator device such as a syringe.

Formulations of the type described above suffer from a number of disadvantages. If the formulation is of low viscosity, it may spread from the  
30 area of application and hence be difficult to apply precisely to the desired area of tissue. If the formulation is more viscous, on the other hand, it may be difficult to dispense. In either case, the formulation, being prepared in

hydrated form, may have a limited lifetime and may be subject to premature curing. It may therefore be necessary for the whole of the formulation to be used at once or discarded. Also, the preparation of formulations immediately prior to use by mixing of ingredients is obviously laborious and time-

5 consuming. In addition to these drawbacks, the degree of adhesion between tissue surfaces that is provided by such formulations may be less than would be desired.

Formulations of tissue adhesive materials have also been applied to a suitable support for application to the tissue surface. The use of therapeutic materials in the form of a sheet, patch or film, for topical administration to either internal or external organs of the body, is well documented for a wide range of medical applications. A disadvantage of products proposed hitherto, however, is that the degree of adhesion to the underlying tissue, particularly in the longer term, may be inadequate. While the initial adhesion may be satisfactory, the sheet may subsequently become detached from the tissue, often after only a few seconds or minutes, eg as a result of hydration of the sheet following its application. In addition, the flexibility of the product may be insufficient for it to conform readily to the surface to which it is applied, which may also have an adverse effect on its adhesion.

As a result of the inadequate adhesion of these products, *inter alia*, it may be necessary to provide further reinforcement, eg through mechanical attachment using sutures, staples or the like. Alternatively, energy (eg light or heat energy) may be applied in order to initiate chemical bonding of the adhesive formulation to the underlying tissue, and hence bonding of the tissue surfaces to each other. Clearly, such approaches introduce further drawbacks. The use of mechanical fastenings such as sutures or staples is often the very thing that the use of such products is intended to replace or avoid. In many instances the use of such fastenings is either not wholly effective (eg on the lung), or undesirable as their introduction gives rise to further areas of tissue weakness. The use of external energy requires the

provision and operation of a source of such energy. Such energy sources may be expensive and difficult to operate, particularly in the confines of an operating theatre or similar environment. Also, the use of external energy for attachment can be both time-consuming and (in some cases) requires  
5 significant careful judgement on the part of the surgeon, to evaluate when sufficient energy has been delivered to effect attachment without damaging the underlying tissue.

There have now been devised improved formulations of tissue-adhesive  
10 materials and sheets or the like of the general type described above that overcome or substantially mitigate the above-mentioned and/or other disadvantages of the prior art.

#### Brief Summary of the Invention

15 According to a first aspect of the invention, there is provided a tissue-adhesive formulation comprising a naturally occurring or synthetic polymerisable and/or cross-linkable material in particulate form, the polymerisable and/or cross-linkable material being in admixture with particulate material comprising  
20 tissue-reactive functional groups.

The formulation according to the invention is advantageous primarily in that it can be easily applied to a tissue surface using a simple applicator or delivery device. As it is applied in solid form, the particulate formulation sticks to the  
25 moist tissue surface and does not spread unduly. On contact with the moist tissue surface the formulation becomes hydrated, thereby causing reaction between the tissue-reactive functional groups and the underlying tissue surface. Reaction may also take place between the tissue-reactive functional groups and the other components of the formulation to form a strong, flexible  
30 and tissue-adherent gel. This formulation thus absorbs physiological fluids (as a consequence of application onto exuding tissue surfaces), combined with additional solutions used to hydrate the formulation following application

(the fluids can be commonly used solutions used in surgical irrigation) becoming gelatinous and adherent to the tissue surfaces, and thereby providing an adhesive sealant, haemostatic and pneumostatic function.

5 In addition, the reaction between the tissue-reactive functional groups and the underlying tissue results in high adhesion between the formulation and the tissue surface, and hence between tissues that are joined using the adhesive formulation. Furthermore, because the formulation is made up in solid form that is, until hydrated by contact with the moist tissue surface (and subsequent  
10 hydration), essentially inactive, the formulation is not prone to premature reaction and as a result its shelf-life may be considerable. This further enables the formulation to be packaged in relatively large quantities that can be dispensed and used over a considerable time period, without the risk of substantial wastage.

15

According to a second aspect of the present invention, there is provided a sheet having a multilayer structure, said structure comprising a core of a naturally occurring or synthetic polymeric material, the core being coated on at least one side thereof with a tissue-adhesive formulation comprising a  
20 naturally occurring or synthetic polymerisable and/or cross-linkable material in particulate form, the polymerisable and/or cross-linkable material being in admixture with particulate material comprising tissue-reactive functional groups.

25 In preferred embodiments of the invention, the tissue-adhesive formulation is applied to the core by mechanically compressing a blend of material containing tissue-reactive functional groups (hereinafter referred to as "tissue-reactive material") and the polymerisable and/or cross-linkable component, both in particulate form, onto one or both sides of the core.

30

The sheet according to the invention is advantageous primarily in that it bonds effectively to tissue, enabling it to be used in a variety of medical applications.

The invention enables coating of the tissue-reactive materials onto (and into) a three-dimensional structural support, whilst maintaining the pliability and physical properties of the support. Furthermore, the adhesive performance of the tissue-reactive materials is not compromised when delivered to the target tissue in this form. Where, as in preferred embodiments, the support is perforated, the perforations provide a means of anchoring the tissue-reactive materials in the support. This reduces or eliminates cracking and crumbling of the tissue-reactive material as it is applied to the support (core), which would result in sub-optimal coverage of the target tissue, and thereby compromise the adhesive/sealant effects of the sheet.

The sheet exhibits good initial adhesion to the tissue to which it is applied (and may thus be described as "self-adhesive"), and furthermore remains well-adhered to the tissue over a longer timescale. Without wishing to be bound by any theory, it is believed that the initial adhesion of the sheet to the tissue is attributable to electronic bonding of the sheet to the tissue, and this is supplemented or replaced by chemical bonding between the tissue-reactive functional groups of the formulation and the tissue, in particular between amine and/or thiol groups on the tissue surface and the tissue-reactive groups of the sheet. Where the structural inner core of the device is perforated, channels can be created (during subsequent hydration following application) that offer a means of escape for fluid that may accumulate at the sheet/material interface. This feature eliminates a common problem associated with the use of adhesives within body cavities, namely over-hydration of the material by exudate, leading to a loss of contact.

The use of the sheet reduces or eliminates the need for additional means of mechanical attachment to the tissue (eg sutures or staples), or the need to provide external energy in the form of heat or light to bring about adherence of the sheet to the underlying tissue. Another advantage of the sheet according to the invention is that it is applied to the tissue as a preformed article, rather than being prepared by mixing of materials immediately prior to use.



By the term "sheet" is meant a three-dimensional article with a thickness that is considerably less than its other dimensions. Such an article may alternatively be described as a patch or a film.

5

According to another aspect of the invention, there is provided a method for the manufacture of a sheet according to the second aspect of the invention, which method comprises forming a core comprising naturally occurring or synthetic polymeric material, and coating at least one side of said core with a tissue-adhesive formulation comprising a naturally occurring or synthetic polymerisable and/or cross-linkable material in particulate form, the polymerisable and/or cross-linkable material being in admixture with particulate material comprising tissue-reactive functional groups.

10

In a third aspect, the invention also provides a method of joining a tissue surface to another tissue, or of sealing a tissue surface, which method comprises applying to the tissue surface a formulation according to the first aspect of the invention or a sheet according to the second aspect of the invention.

20

#### Brief Description of the Drawings

Figure 1 is a schematic representation of the reaction between a tissue-reactive functional group (in the illustrated case an N-hydroxysuccinimide ester) and an amine-containing molecule such as a tissue protein.

25

Figure 2 shows the introduction of carboxyl group-bearing side chains into a poly(vinyl alcohol - vinyl acetate) copolymer.

Figure 3 represents the formation of a poly(N-vinyl-2-pyrrolidone-co-acrylic acid) copolymer.

30

Figure 4 shows the mechanism of free radical initiation of a polymerisation reaction.

Figure 5 illustrates the synthesis of a tissue reactive material.

5

Figure 6 is a schematic sectional view of a sheet according to the invention.

### Detailed Description of the Invention

#### 10 *Composition of the coating formulation*

The formulation with which the core is coated comprises a naturally occurring or synthetic polymerisable and/or cross-linkable material in particulate form, and a particulate material comprising tissue-reactive functional groups.

15

These two components may be blended in suitable proportions, which may depend on the particular materials used, as well as the desired properties of the resulting blend. Typically, the ratio of cross-linkable material to material comprising tissue-reactive functional groups will be between 0.2:1 and 10:1,

20 more preferably between 0.5:1 and 10:1.

#### *Nature of the particulate material comprising tissue-reactive groups*

The tissue-reactive material is preferably polymeric in nature. Most  
25 preferably, the polymer is a synthetic polymer.

By "tissue-reactive functional groups" is meant functional groups capable of reacting with other functional groups present in the tissue surface so as to form covalent bonds between the formulation and the tissue. Tissues  
30 generally consist partly of proteins, which commonly contain thiol and primary amine moieties. Many functional groups such as imido ester, p-nitrophenyl carbonate, N-hydroxysuccinimide (NHS) ester, epoxide, isocyanate, acrylate,

vinyl sulfone, orthopyridyl-disulfide, maleimide, aldehyde, iodoacetamide, and others, may react with thiols or primary amines, and therefore constitute "tissue-reactive functional groups".

- 5 Figure 1 illustrates the mechanism by which an NHS-functionalised polymer reacts with an amine-containing material such as a tissue protein represented by  $R-NH_2$ . The reaction is a nucleophilic displacement leading to the formation of an amide bond between the polymer and the tissue protein.
- 10 Tissue-reactive functional groups that may be of utility in the present invention are any functional groups capable of reaction (under the conditions prevalent when the formulation is applied to tissue, ie in an aqueous environment and without the application of significant amounts of heat or other external energy) with functional groups present at the surface of the tissue. The latter class of
- 15 functional group includes thiol and amine groups, and tissue-reactive functional groups therefore include groups reactive to thiol and/or amine groups. Examples are:
- imido ester
  - p-nitrophenyl carbonate
  - 20 N-hydroxysuccinimide (NHS) ester
  - epoxide
  - isocyanate
  - acrylate
  - vinyl sulfone
  - 25 orthopyridyl-disulfide
  - maleimide
  - aldehyde
  - iodoacetamide
- 30 N-hydroxysuccinimide (NHS) ester is a particularly preferred tissue-reactive functional group.

In general, the tissue-reactive material may be formed by derivatisation of a suitable polymer precursor. Classes of polymer which lend themselves to such derivatisation include those that contain carboxylic acid or alcohol functional groups, or related structures. Polymers that may be used include

5 polymers that are commercially available or polymers that are prepared specifically for this purpose. Naturally-occurring materials such as sucrose or a derivatised cellulose may also be used.

Commercially available polymers that may be used include polyvinylalcohol (PVA). In the case of PVA, the functional groups may be introduced by first

10 adding a chain extending or linking group, for example an acid functionality that can be further reacted with N-hydroxy succinimide. Figure 2 shows the addition of a chain-extending group to a copolymer of vinyl acetate and vinyl alcohol, the chain-extending group terminating in a carboxylic acid group that

15 may be converted to the corresponding NHS-ester. The copolymer starting material (in which molar fraction  $x$  of vinyl alcohol groups may be 0.85-0.995) is reacted with a cyclic anhydride (in the example illustrated, succinic anhydride) in the presence of a base such as pyridine. Between 5% and 40% of the alcohol groups are derivatised to form the carboxylic acid-bearing side

20 chains (ie  $a+b=x$ , with  $a$  being between  $0.05x$  and  $0.40x$ ), which may then be converted to the NHS-ester by conventional methods that are known *per se*.

Where the polymer support is synthesized for the purpose of subsequent derivatization, a wide variety of monomers may be used. Examples include

25 N-vinyl-2-pyrrolidone, acrylic acid, vinyl acetate, vinyl acetic acid, mono-2-(methacryloyloxy)ethyl succinate, methacrylic acid, 2-hydroxyethyl methacrylate, 2-hydroxypropyl methacrylate, (polyethylene glycol) methacrylate or other monomers containing acid or alcohol functionality. Such monomers may be polymerised via various standard polymerisation

30 techniques, including free radical techniques using an initiator such as benzoyl peroxide, 2,2'-azobisisobutyronitrile (AIBN), lauroyl peroxide, peracetic acid etc. One preferred example of such a polymer is poly(N-vinyl-

2-pyrrolidone-co-acrylic acid) co-polymer polymerised using AIBN as initiator.

The polymerization of this material is illustrated in Figure 3, in which the molar ratio of acrylic acid-derived units may be between 0 and 1.0, preferably less than 0.60, and most preferably between 0.025 and 0.25. The copolymer may be further reacted with N-hydroxysuccinimide to form the tissue-reactive material.

Where, as is preferred, the tissue-reactive functional groups are NHS-esters, at least one of the monomers used in the preparation of the tissue-reactive material must contain a carboxylic acid group or a group capable of being reacted with another material to form an acid functionality.

In the preferred case in which the tissue-reactive material is an NHS ester of poly(N-vinyl-2-pyrrolidone-co-acrylic acid) copolymer (PVP-co-PAA), the molar ratio of acrylic acid-derived units is preferably between 0.05 and 0.50, and hence that of the vinyl pyrrolidone-derived units is between 0.50 and 0.95.

The acrylic acid groups are preferably derivatised with NHS groups. A copolymer of PVP and PAA, in which the carboxyl groups of the acrylic acid-derived units carry NHS groups, is referred to herein as NHS-activated PVP-co-PAA.

As noted above, the activity of the tissue-reactive material (ie the degree to which the tissue-reactive functional groups of that material bind to the tissue) may be controlled by varying the proportion of that material in the formulation. The concentration of the tissue-reactive material in the formulation may be varied quite widely, eg from 10% w/w or less up to 50% w/w or more.

The formulation may contain one type of tissue-reactive material, or more than one type of tissue-reactive material.

Preferably, all or substantially all of the available sites in the precursor to the tissue-reactive material will be derivatised (ie the tissue-reactive functional groups will be introduced into all or substantially all of the available sites in the precursor to the tissue-reactive material). The degree of binding between the tissue-reactive functional groups and the tissue to which the formulation is applied will then be a function of the amount of tissue-reactive material in the formulation.

*Nature of the polymerisable and/or cross-linkable component*

The polymerisable and/or cross-linkable component of the formulation is preferably selected from polysaccharides, polylactates, polyols and proteins, and derivatives thereof.

The polymerisable component may be in a crosslinked form in which individual molecules of the polymerisable component are linked together through intermolecular covalent bonds. Such crosslinking can be effected by standard techniques known in the art, for example by heat treatment and/or crosslinking agents. Depending on the nature of the polymerisable component and/or the conditions employed to effect crosslinking, the degree of crosslinking between individual molecules can vary considerably.

The degree of pre-crosslinking in the polymerisable components, however, should not be such that it substantially prevents the subsequent reaction of the tissue reactive functional groups with the polymerisable component.

Proteins are preferred materials for the polymerisable and/or cross-linkable component of the formulation because they are rich in functional groups that are reactive to tissue-reactive functional groups. Hence, the tissue-reactive functional groups will react not only with the tissue surface to which the formulation is applied, but also with the polymerisable and/or cross-linkable component of the formulation.

A particularly preferred protein for use in the invention is albumin, particularly mammalian albumin such as porcine, bovine or human albumin.

- 5 Preferred synthetic polymers that may be, or may be present in, the polymerisable and/or cross-linkable component of the formulation include multifunctionally activated synthetic polymers, ie synthetic polymers that have, or have been chemically modified to have, a plurality of functional groups that are capable of reacting with each other or with other functional groups present
- 10 in the formulation to form covalent bonds. Preferred multifunctionally activated synthetic polymers include chemically modified polyalkylene glycols, in particular polyalkylene glycols that have been modified to contain multiple primary amino or thiol groups.
- 15 Suitable modified polyalkylene glycols include both linear and branched ((et so-called "3-arm" and "4-arm") compounds. Examples of suitable multi-amino polymers are those sold under the trade name JEFFAMINE. These are based on backbones of polyethylene glycol and/or polypropylene glycol units with terminal amine groups. Methods by which similar and analogous chemically
- 20 modified polymers can be prepared will be readily apparent to those skilled in the art.

#### *Manufacture of the components of the formulation*

- 25 The particulate tissue-reactive material and the particulate polymerisable and/or cross-linkable component may be prepared by any suitable means. Particularly where the latter component is proteinaceous, the particles are preferably prepared by freeze- or heat-drying of an aqueous solution or suspension. To enhance subsequent reaction of the proteinaceous material
- 30 with the tissue-reactive material, the solution or suspension is preferably buffered to an alkaline pH prior to drying.

The formulation may be prepared simply by admixing the components in particulate form, and where desired compacting the formulation to form tablets, plugs or the like. The degree of compaction should be such that the tablets etc retain their integrity until applied to tissue, but not so great as to inhibit hydration (and hence adhesion) after application.

*Physical forms of the formulation*

The formulation according to the invention may have the form of a loose powder, in which particles of the tissue-reactive material are admixed with particles of the polymerisable and/or cross-linkable component.

Alternatively, the formulation may take the form of a compacted body formed by compaction of the particles. Tissue-reactive materials based on poly (N-vinyl-2-pyrrolidone) or copolymers of N-vinyl-2-pyrrolidone with other monomers (eg vinylic monomers) are particularly preferred in such applications, as poly (N-vinyl pyrrolidone) has suitable flow properties for blending with other components of the formulation, and exhibits excellent performance in dry granulation tableting processes as it undergoes plastic deformation on compression, and has low hygroscopicity.

In a further alternative, the formulation may be applied to a core to form a sheet according to the second aspect of the invention.

The formulation may further comprise additional components such as structural polymers, surfactants, plasticizers and excipients commonly used in tablet manufacture. Such further components may be present as discrete particles, or may be components of the particles of tissue-reactive material and/or polymerisable and/or cross-linkable component.

*Nature of the core*



The principal functions of the core are to provide the sheet with structural integrity and to provide a flexible substrate onto which the tissue-reactive formulation, in powder form, can be applied.

- 5 The core can be prepared using any suitable polymeric material or combination of materials. The core may be biodegradable or non-biodegradable, and should be biocompatible, ie should be capable of application to tissues either within or external to the body without causing any immunological or other adverse reaction.

10

Examples of polymeric materials that may be used for the core are:

Polymers or co-polymers based on  $\alpha$ -hydroxy acids, such as polylactide, polyglycolide, and also polycaprolactone and other polylactones such as

15

Other examples may include:

Alginates (ie polymers based on alginic acid, the polysaccharide obtained

20

Polyhydroxyalkanoates

Polyamides

Polyethylene

Propylene glycol

25

Water-soluble glass fibre

Starch

Cellulose

Collagen

Pericardium

30

Albumin

Dacron

Polyurethane

Polypropylene

Polytetrafluoroethylene

5 The core may be prepared by casting, spinning or foaming of a solution of the polymeric material, or by moulding, weaving of filamentous material, or slicing from a block of material. Appropriate techniques for the preparation of the core by such methods will be familiar to those skilled in the art.

10 In preferred embodiments, the core is formed with a regular array of apertures; for example in a square or hexagonal array. The apertures may be formed during manufacture of the core or may be introduced after the core is formed, for example by piercing.

15 Preferably, the apertures are between  $50\mu\text{m}$  and  $2\text{mm}$  in diameter and adjacent apertures are formed at a centre-to-centre separation of between  $100\mu\text{m}$  and  $5\text{mm}$ . Preferably, the apertures account for between 5% and 80% of the overall surface area of the core.

20 The core may have a thickness of from  $0.005$  to  $5\text{mm}$ .

*Application of the formulation to the core*

25 The formulation may be applied to just one side of the core. More preferably, however, the formulation is applied to both sides of the core.

30 Where the core is apertured, application of the formulation to one or both sides of the core causes the apertures to be filled with the formulation. In use, where the formulation is applied to both sides of an apertured core, the formulation that is present in the perforations effectively binds together the activated coatings on each side of the core, encapsulating the core between the two layers of activated coating:

The preferred method for applying the formulation to the core involves mechanically compressing (eg using a hydraulic press) a blend of the tissue-reactive material and the polymerisable and/or cross-linkable component, both in particulate form, onto one or both sides of the core.

The blend may be prepared by admixing particles of the tissue-reactive material with particles of the polymerisable and/or cross-linkable component.

The coating formulation may include filler materials that may typically account for up to 50% by weight of the coating formulation. Examples of such materials include cellulose derivatives (eg carboxymethyl cellulose, hydroxypropyl methylcellulose, etc), polyethylene glycol, polyvinylpyrrolidone and other commonly used pharmaceutical excipients.

The thickness of the coating applied to one or both sides of the core will typically be between 50µm and 0.5mm, more commonly from about 70µm to about 0.2mm.

Optionally, a surface of the sheet that, in use, is not intended to adhere to tissue may be coated with a non-adhesive material. Most preferably, such a material is a synthetic polymer. Examples of suitable polymers include polyethylene glycols, polylactide and poly(lactide-co-glycolide). A sheet with such a non-adhesive coating will adhere only to the target tissue (to which the underside of the sheet is applied) and not to surrounding tissues (eg the pleural or peritoneal wall). The non-adhesive coating may include a visibly-absorbing chromophore to enable identification of the non-tissue contacting surface of the sheet. An example of a suitable chromophore is methylthioninium chloride.

*Physical form of the sheet*

The sheet may typically have an overall thickness of from 0.05 to 10 mm, typically 0.05 to 2mm, and more commonly 0.05 to 0.5 mm, eg about 200µm or 300µm or 400µm.

- 5 The sheet may be produced with, or subsequently cut to, dimensions of from a few square millimetres up to several tens of square centimetres.

*Therapeutic applications of the formulation and sheet*

- 10 The formulation and sheet according to the invention are suitable for application to both internal and external surfaces of the body, ie they may be applied topically to the exterior of the body (eg to the skin) or to internal surfaces such as surfaces of internal organs exposed during surgical procedures.

15

The formulation and sheet according to the invention are particularly suitable for surgical applications in the following areas:

Thoracic / cardiovascular

- 20 General surgery

ENT

Urology

Oral / maxillofacial

Orthopaedic

- 25 Neurological

Gastroenterology

Ophthalmology

Gynaecology / obstetrics

- 30 Possible uses are described in more detail below.

### *Wound healing*

The formulation and degradable nature of the sheet mean that they may support and promote wound healing during both internal and topical procedures. Once the formulation and/or sheet begin to degrade, fibroblasts will move in and begin to deposit components of the extracellular matrix. The formulation and sheet therefore can be used as an internal or external dressing. In addition, factors such as growth factors and cAMP that are known to promote the proliferation of skin cells may be added to the formulation to assist in the healing process. The sheet may be designed to control the transmission of moisture and infectious agents, and thus be useful particularly in the treatment of burns.

### *Skin closure*

The formulation and sheet may be applied topically to promote wound closure (as an alternative to sutures). This may have beneficial effects in that it may reduce scarring, and the formulation and sheet may thus be useful for cosmetic purposes during minor surgery (eg in Accident and Emergency Departments). The self-adhesive properties of the sheet make it easy to apply quickly.

### *Hernia repair*

The sheet may be used to provide reinforcement in hernia repair procedures. The self-adhesive attachment overcomes the potential issues faced by conventional surgical reinforcing mesh products, which require suturing or stapling in an already weakened area. The sheet for such a procedure may be engineered to have short or long term durability, depending on the degree of tissue repair required. The sheet may also be able to withstand the application of staples.

### *Anastomosis*

The formulation and self-adhesive sheet provide a mean of rapid sealing of, and prevention of leaks in, joined tubular structures such as blood vessels,

and vascular and bladder grafts, and the GI tract. The ability of the sheet to support tissue repair may be of particular value if used in nerve repair.

#### *Sealing large areas of tissue*

- 5 The good sealing and handling properties of the formulation and sheet, combined with their self-adhesive properties and ability to cover a large surface area, mean that they may be of particular use in sealing resected tissue surfaces – in particular those where diffuse bleeding is an issue (eg the liver). The sheet also provides an ideal support matrix for tissue repair at  
10 such sites. This could also be applicable to limiting leakage of cerebro-spinal fluid following neurological surgery.

#### *Sealing air leaks*

- In addition to the patch properties described above, the high tensile strength  
15 and good inherent elasticity of the formulation and sheet (after hydration and reaction of the tissue-reactive functional groups), make them particularly suitable for sealing air leaks in the lung, particularly following lung resection. Again, after effecting a seal, the sheet provides an ideal support matrix for tissue repair at such sites.

20

#### *Haemostasis*

- The formulation and sheet may be applied to a bleeding area, acting as a physical barrier. The tissue-reactive material in the formulation and sheet may immobilise proteins and thereby promote haemostasis.

25

#### *Therapeutic agent administration*

- Drugs and other therapeutic agents (including biologically active agents such as growth factors, and even cells and cellular components) may be added to solution(s) used to form the components of the formulation and sheet, or  
30 covalently linked to components prior to their use in the manufacture of the formulation and sheet. Once the formulation or sheet is in place, following application to the desired site, the drug will be slowly released, either by

diffusion or by engineering the formulation or sheet so that as it degrades over time the drug is released. The rate of release can be controlled by appropriate design of the formulation and sheet. The formulation and sheet may thus provide a mean of delivering a known amount of drug either systemically or to a precise locus. The drug may be directly bound to a component of the formulation, or simply dispersed in the formulation.

#### *Prevention of Post-Surgical Adhesions*

Post-surgical adhesion, the formation of undesired connective tissue between adjacent tissues, is a serious problem which can give rise to major post-surgical complications. It is a particular problem in bowel surgery where it can cause, for instance, twisting of the bowel, which may then necessitate further surgical intervention. The application of sheet material having self-adhesive properties in accordance with the invention to tissues exposed in a surgical procedure can be effective in preventing post-surgical adhesions between that tissue and neighbouring tissues.

#### Detailed Description of Preferred Embodiments

The invention will now be described in greater detail, by way of illustration only, with reference to the following Examples.

##### Example 1

##### Synthesis of NHS-activated PVP-co-PAA

##### (a) Polymerisation of acrylic acid and N-vinyl-2-pyrrolidone

The polymer is formed via the polymerisation of monomers such as N-vinyl-2-pyrrolidone and acrylic acid, as shown in Figure 3.

A number of methods may be used to initiate the polymerisation, such as free radical, ionic (cationic or anionic), thermal, UV, redox etc. Free radical

polymerisation is the preferred polymerisation method and 2-2'-azo-bis-isobutyronitrile (AIBN) is the preferred initiator. The AIBN decomposes into two radicals which can then attack the carbon-carbon double bond in the vinylic monomer (acrylic acid) as shown in Figure 4.

5

This will continue until termination of chain growth, via combination, disproportionation etc.

10

The reaction solvent may be N,N'-dimethylformamide, toluene, or any other suitable solvent with a boiling point greater than 100°C. Toluene is the currently preferred solvent.

A typical polymerisation method is as follows:

15

Solvent is charged to the reaction flask, usually around 5-10ml of solvent per gram of monomer used is sufficient. The solvent is heated in an oil bath to a temperature sufficient for the generation of free radicals from the chosen initiator, 80-85°C is the optimum temperature when using AIBN as the initiator. Oxygen-free nitrogen is bubbled through the solvent to remove any dissolved oxygen. Oxygen is also removed from the monomers in the same manner. The initiator is added to the solvent and allowed to dissolve. The monomers are added and the vessel closed. A nitrogen inlet and an escape needle may also be used.

20

25

The reaction is allowed to stand for around 18 hours. The reaction mixture is cooled and the polymer is isolated from the solvent/polymer solution by precipitation in 5:1 hexane/isopropanol followed by filtration. Successive washes with diethyl ether are required to remove all traces of polymerisation solvent from the polymer. After 4-5 diethyl ether washes the polymer is dried under reduced pressure to constant weight.

30

Typical reaction conditions are shown in Table I:



Table I

Solvent (vol)	Monomer (g)		AIBN (g)	T (°C)	Time (hrs)	Yield	Mn	Mw	Polydispersity Index
	acrylic acid	N-vinyl-2- pyrrolidone							
Toluene (100ml)	1.5 (20mol%)	8.5 (80mol%)	0.02 (0.125%)	80	3	-	-	-	-
Toluene (100ml)	0.7 (10mol%)	9.3 (90mol%)	0.02 (0.125%)	80	3	54%	80040	38800	2.0
Toluene (100ml)	0.7 (10mol%)	9.3 (90mol%)	0.04 (0.25%)	80	3	58%	74240	38340	1.9
DMF (100ml)	0.7 (10mol%)	9.3 (90mol%)	0.02 (0.125%)	80	3	62%	54000	25150	2.1
Toluene (100ml)	0.5 (7.5mol%)	9.5 (92.5mol%)	0.02 (0.125%)	80	3	-	-	-	-
Toluene (100ml)	0.35 (5mol%)	9.65 (95mol%)	0.2 (0.125%)	80	3	-	-	-	-

(b) Reaction of PVP-co-PAA and N-hydroxysuccinimide in the presence of dicyclohexylcarbodiimide

5 NHS-activated PVP-co-PAA is formed from the reaction of PVP-co-PAA and N-hydroxysuccinimide in the presence of dicyclohexylcarbodiimide (Figure 5).

10 10g of PVP-co-PAA containing 0.094 moles of acrylic acid repeat units is dissolved in 50 ml of dried N,N'-dimethylformamide by stirring in a dry 100ml round bottomed flask. 0.01 moles of N-hydroxysuccinimide (1.15g) is added to the polymer solution and is allowed to dissolve.

15 Dicyclohexylcarbodiimide (2.06g) is melted in an oven at 60°C and added to the polymer solution. This is left to stir at room temperature for at least 24 hours. The formation of a white precipitate (dicyclohexylurea) is observed. After 24 hours the precipitate is removed by filtration, and the flask and filter washed with a small amount of dry DMF. The polymer is isolated by precipitation in 5:1 hexane/isopropanol and filtration. The polymer is further purified by repeated washes with dry diethyl ether. The yield is between 50-70%.

20 Example 2

Blending of NHS-activated PVP-co-PAA with freeze-dried albumin

25 a) Powders of NHS-activated PVP<sub>80</sub>-co-PAA<sub>20</sub> copolymers have been blended (1:1) with freeze dried porcine albumin (Sigma Aldrich; previously buffered to pH 10.5).

b) Powders of NHS-activated PVP<sub>70</sub>-co-PAA<sub>30</sub> copolymers have been blended (1:1) with freeze-dried human albumin (Baxter human albumin infusion (20%) previously buffered to pH 10.5 buffer).

30 c) Powders of NHS-activated PVP<sub>70</sub>-co-PAA<sub>30</sub> copolymers have been blended (2:1) with freeze dried porcine albumin (previously buffered to pH 10.5 buffer).

Example 3Blending of NHS-activated PVP-co-PAA with freeze-dried albumin and application to liver tissue

- 5 a) Powders of NHS-activated PVP<sub>80</sub>-co-PAA<sub>20</sub> copolymers have been blended (1:1) with freeze dried porcine albumin (Sigma Aldrich; previously buffered to pH 10.5) and delivered onto moist liver tissue. The powder rehydrated rapidly (< 5 minutes) yielding a gel that offers cohesive strength, in addition to offering strong adhesion to the underlying tissue surface.
- 10
- b) Powders of NHS-activated PVP<sub>70</sub>-co-PAA<sub>30</sub> copolymers have been blended (1:1) with freeze-dried human albumin (Baxter human albumin infusion (20%) previously buffered to pH 10.5 buffer) and delivered onto moist liver tissue. The powder rehydrated rapidly (< 5 minutes) yielding a gel that offers cohesive strength, in addition to offering strong adhesion to the underlying tissue surface.
- 15

Example 4Blending of NHS-activated PVP-co-PAA with freeze-dried porcine albumin, forming a compressed disc followed by application to liver tissue

- Powders of NHS-activated PVP<sub>70</sub>-co-PAA<sub>30</sub> copolymers have been blended (2:1) with freeze dried porcine albumin (previously buffered to pH 10.5 buffer), followed by compression into a thin (<2mm thick) disc and delivered onto moist liver tissue. The disc adheres immediately to the liver tissue and rehydrates gradually over an hour yielding a gel that offers cohesive strength, in addition to offering strong adhesion to the underlying tissue surface.
- 25

Example 5Blending of excipients with powdered PVP(NHS)<sub>20</sub> and freeze-dried porcine albumin previously buffered to pH 10.5 (PSA)

Powders of PVP(NHS)<sub>20</sub> and PSA (1:1) have been blended with excipients such as hydroxypropyl cellulose, poly(vinyl pyrrolidone) and microcrystalline cellulose. The powdered mixture was compressed into a disc with a thickness of less than 2mm. These discs adhered immediately to moist porcine liver tissue and rehydrated upon immersion in aqueous solution. After immersion in aqueous solution for 1 hour, they remained adhered to tissue as a crosslinked gel.

Adhesion was obtained with concentrations of PVP(NHS) and PSA from 11.5% to 50% w/w.

#### 10 Example 6

##### Preparation of perforated poly(lactide-co-glycolide) (PLG) film

A PLG film, approx 50-100µm thick is placed between two blocks of perspex or other sheet material. The perspex blocks have a series of pre-drilled holes in registration with each other. A syringe needle or the like is pushed through the holes in the perspex blocks, thereby puncturing holes in the PLG film. The needle is withdrawn, leaving a hole in the film. The process is repeated until a series of holes of equal diameter and equal spacing have been punctured in the film. The film is released from between the two blocks of perspex.

#### 20 Example 7

##### Application of coating formulation to the film

The perforated PLG film prepared in accordance with Example 6 is cut to the desired shape using a suitable 13mm diameter punch. Into a suitable tablet die, 15mg of the PVP(NHS):HSA co-powder (4:1) is loaded evenly onto the bottom surface ensuring an even distribution of material. The pre-cut perforated PLG film is placed on top of this semi-compressed powder layer ensuring close contact between the film and the powder beneath. Another layer of powder (15mg; same composition as the first layer) is loaded onto the PLG film and distributed evenly. The tablet die is loaded into a hydraulic press and the film/powder composite is compressed at a load of less than 0.5 tonne.

### Example 8

#### Application of sheet to tissue

- 5 The tissue surface is prepared in accordance with conventional surgical technique. The sheet is applied onto (and if necessary) around the tissue surface with moderate pressure to ensure satisfactory contact to the tissue. Following application, the sheet is hydrated with saline.

### Example 9

#### Measurement of Adhesive Strength

- 10 A Universal testing machine (UTM, Zwick/Roell BZ2.5) was used to test the adhesive strength of test materials to freshly excised liver or lung tissue. Details of the testing procedure are summarised below.

- A small section of tissue (4 cm × 4 cm × 1 cm (depth)) was prepared and mounted into a purpose-made holder at the base of the test machine. The surface of the tissue was sprayed with saline. The test specimen (with sample holder attached to enable subsequent removal) was placed onto the tissue surface with a moderate force to ensure full contact. The material was left on the tissue for 5 minutes and then wholly submerged in saline for a further 5 minutes. Whilst holding the tissue in place using a suitable clamp the folded tip of the sample holder was inserted in the grips of the UTM. The sample was positioned appropriately to ensure that the sample was aligned with the grips. The grip was then moved at 180° from the test sample thereby removing the sample from the tissue. The UTM software (Zwick TestXpert ver 9.0) can be used to calculate the energy of adhesion (mJ) of the test material. Adhesion tests were performed on powder compositions of differing compositions and compared to a commercially available polyethylene glycol (PEG) based sealant.

Table II shows the data obtained by testing compressed films as a function of composition.

Table II

5

Test medium Substrate	Tissue reactive material	Crosslinkable material	Ratio of components	Mean energy of adhesion (SD)/ mJ' (n=6)
Porcine liver	PVP(NHS)	Human albumin	4:1	1.3 (0.29)
Porcine liver	PVP(NHS)	Human albumin	1:4	1.0 (0.43)
Porcine liver	PVP(NHS)	Human albumin	1:1	1.0 (0.27)

The results listed in Table II demonstrate the adhesion performance of the co-powder formulations of PVP(NHS)<sub>20</sub> and freeze dried human albumin.

- 10 In a further study, Table III illustrates the results of adhesion testing of sheet formulations, (13mm diameter circular discs) studied as a function of the number of apertures in a PLG film with active co-powders pressed into either side of the PLG film. The powders utilised were PVP(NHS)<sub>20</sub> and human albumin in a 4:1 ratio.

15

Table III

Test medium	No of apertures in 13mm diameter sheet	Mean energy of adhesion (SD)/ mJ (n=6)
Porcine liver	0	0.39 (0.14)
Porcine liver	10	0.75 (0.25)
Porcine liver	20	1.14 (0.37)

The results listed in Table III demonstrate that the energy of adhesion of multi layered sheets is proportional to the number of apertures in the PLG inner film.

Example 10

5 Schematic representation of a sheet according to the invention

Figure 6 shows (schematically and not to scale) the structure of a typical sheet prepared in accordance with the invention: The sheet comprises a core in the form of a film 1 of PLG which has a regular array of apertures. Layers 2,3 of a  
10 tissue-reactive formulation are pressed onto both sides of the film 1 such that the tissue-reactive formulation penetrates into, and fills, the apertures. Finally, a non-adhesive layer 4, again of PLG, is applied to one surface of the sheet. The non-adhesive layer 4 may include a chromophore that gives the non-adhesive surface a discernible colour, thereby identifying that surface (and hence indicating which  
15 side of the sheet is to be applied to the tissue).

**Claims**

1. A tissue-adhesive formulation consisting of a naturally occurring or synthetic polymerisable and/or cross-linkable material in particulate form, the polymerisable and/or cross-linkable material being in admixture with particulate material comprising tissue-reactive functional groups.
2. A formulation according to Claim 1, wherein the ratio of polymerisable and/or cross-linkable material to material comprising tissue-reactive functional groups is between 0.2:1 and 10:1.
3. A formulation according to Claim 2, wherein the ratio of polymerisable and/or cross-linkable material to material comprising tissue-reactive functional groups is between 0.5:1 and 10:1.
4. A formulation according to any preceding claim, wherein the tissue-reactive functional groups are selected from the group consisting of imido ester, p-nitrophenyl carbonate, N-hydroxysuccinimide (NHS) ester, epoxide, isocyanate, acrylate, vinyl sulfone, orthopyridyl-disulfide, maleimide, aldehyde and iodoacetamide.
5. A formulation according to Claim 4, wherein the tissue-reactive functional groups are N-hydroxysuccinimide (NHS) esters.
6. A formulation according to any preceding claim, wherein the formulation contains one type of material comprising tissue-reactive functional groups.
7. A formulation according to any one of Claims 1-5, wherein the formulation contains two types of materials comprising tissue-reactive functional groups.
8. A formulation according to any preceding claim, wherein the material comprising tissue-reactive functional groups is formed by derivatization of a polymer precursor.



9. A formulation according to Claim 8, wherein all or substantially all of the available sites in the polymer precursor are derivatised.
- 5 10. A formulation according to Claim 8 or Claim 9, wherein the polymer precursor contains carboxylic acid or alcohol functional groups.
- 10 11. A formulation according to Claim 10, wherein the polymer precursor is selected from the group consisting of sucrose, cellulose and polyvinylalcohol (PVA).
- 15 12. A formulation according to Claim 10, wherein the polymer precursor is formed by polymerisation of two or more monomers, and at least one of the monomers contains a carboxylic acid group or a group capable of being reacted with another material to form an acid functionality.
- 20 13. A formulation according to Claim 12, wherein the monomers are selected from the group consisting of *N*-vinyl-2-pyrrolidone, acrylic acid, vinyl acetate, vinyl acetic acid, mono-2-(methacryloyloxy)ethyl succinate, methacrylic acid, 2-hydroxyethyl methacrylate, 2-hydroxypropyl methacrylate and (polyethylene glycol) methacrylate.
- 25 14. A formulation according to Claim 12 or Claim 13, wherein polymerisation is initiated by a free radical initiator.
- 30 15. A formulation according to Claim 14, wherein the initiator is selected from the group consisting of benzoyl peroxide, 2,2'-azobisisobutyronitrile (AIBN), lauroyl peroxide and peracetic acid.
16. A formulation according to any one of Claims 12 to 15, wherein the polymer precursor is poly(*N*-vinyl-2-pyrrolidone-co-acrylic acid) co-polymer.

17. A formulation according to Claim 16, wherein the poly(N-vinyl-2-pyrrolidone-co-acrylic acid) co-polymer has a molar ratio of acrylic acid-derived units less than 0.60.
- 5 18. A formulation according to Claim 16, wherein the poly(N-vinyl-2-pyrrolidone-co-acrylic acid) co-polymer has a molar ratio of acrylic acid-derived units between 0.025 and 0.25.
- 10 19. A formulation according to any one of Claims 7 to 18, wherein the polymer precursor is reacted/derivatized with N-hydroxysuccinimide to form the material comprising tissue-reactive functional groups.
- 15 20. A formulation according to Claim 19, wherein the material comprising tissue-reactive functional groups is an N-hydroxysuccinimide ester of poly(N-vinyl-2-pyrrolidone-co-acrylic acid) co-polymer.
- 20 21. A formulation according to Claim 20, wherein the material comprising tissue-reactive functional groups has a molar ratio of acrylic acid-derived units between 0.05 and 0.50 and vinyl pyrrolidone-derived units between 0.50 and 0.95.
22. A formulation according to any preceding claim, wherein the concentration of material comprising tissue-reactive functional groups in the formulation is between 10 and 50% w/w.
- 25 23. A formulation according to any preceding claim, wherein the polymerisable and/or cross-linkable material is selected from the group consisting of polysaccharides, polylactates, polyols and proteins, and derivatives thereof.
- 30 24. A formulation according to any one of Claims 1 to 22, wherein the polymerisable and/or cross-linkable material is, or further comprises, a chemically modified polyalkylene glycol containing multiple primary amino or thiol groups.

25. A formulation according to Claim 24, wherein the chemically modified polyalkylene glycol is that sold under the trade name JEFFAMINE.
- 
26. A formulation according to Claim 23, wherein the polymerisable and/or cross-linkable material is cross-linked.
27. A formulation according to Claim 23 or Claim 26, wherein the polymerisable and/or cross-linkable material is albumin.
28. A formulation according to Claim 27, wherein the polymerisable and/or cross-linkable material is porcine, bovine or human albumin.
29. A formulation according to any preceding claim, further comprising one or more further components selected from structural polymers, surfactants, plasticisers and other excipients.
30. A sheet having a multilayer structure, said structure consisting of a core of a naturally occurring or synthetic polymeric material, the core being coated on at least one side thereof with a tissue-adhesive formulation according to any preceding claim.
31. A sheet according to Claim 30, wherein the core is biocompatible with the tissue surface to which it is applied.
32. A sheet according to Claim 31, wherein the core comprises polymeric material selected from the group consisting of polymers or co-polymers based on  $\alpha$ -hydroxy acids such as polylactide, polyglycolide, polycaprolactone and other polylactones such as butyro- and valerolactone.
33. A sheet according to Claim 31, wherein the core comprises polymeric material selected from the group consisting of alginates, polyhydroxyalkanoates, polyamides, polyethylene, propylene glycol, water-soluble glass fibre, starch,

cellulose, collagen, pericardium, albumin, Dacron, polyurethane, polypropylene and polytetrafluoroethylene.

- 5 34. A sheet according to any one of Claims 30 to 33, wherein the core is apertured.
- 10 35. A sheet according to Claim 34, wherein the sheet has a regular arrays of apertures, and the apertures are between 50 $\mu$ m and 2mm in diameter and adjacent apertures are formed at a centre-to-centre separation of between 100 $\mu$ m and 5mm.
36. A sheet according to Claim 35, wherein the apertures account for between 5% and 80% of the overall surface area of the core.
- 15 37. A sheet according to any one of Claims 30 to 36, wherein the core has a thickness of 0.005 to 5mm.
- 20 38. A sheet according to any one of Claims 30 to 37, wherein the tissue-adhesive formulation is applied to the core by mechanically compressing a blend of material containing tissue-reactive functional groups and polymerisable and/or cross-linkable material, both in particulate form, onto one or both sides of the core.
- 25 39. A sheet according to Claim 38, wherein the core is coated on both sides with the said blend of material.
40. A sheet according to Claim 39, wherein one surface of the sheet is coated with a non-adhesive material.
- 30 41. A sheet according to Claim 40, wherein the non-adhesive material is selected from the group consisting of polyethylene glycols, polylactide and poly(lactide-co-glycolide).

42. A sheet according to Claim 41, wherein the non-adhesive coating includes a visibly-absorbing chromophore.

43. A sheet according to Claim 42, wherein the visibly-absorbing chromophore is methylthioninium chloride.

44. A method for the manufacture of a sheet according any one of Claims 30 to 43, which method comprises forming a core consisting of naturally occurring or synthetic polymeric material, and coating at least one side of said core with a tissue-adhesive formulation consisting of a naturally occurring or synthetic polymerisable and/or cross-linkable material in particulate form, the polymerisable and/or cross-linkable material being in admixture with particulate material consisting of tissue-reactive functional groups.

45. A method of joining a tissue surface to another tissue, or of sealing a tissue surface, which method comprises applying to the tissue surface a formulation according to any one of Claims 1 to 29 or a sheet according to any one of Claims 30 to 43.

Figure 1

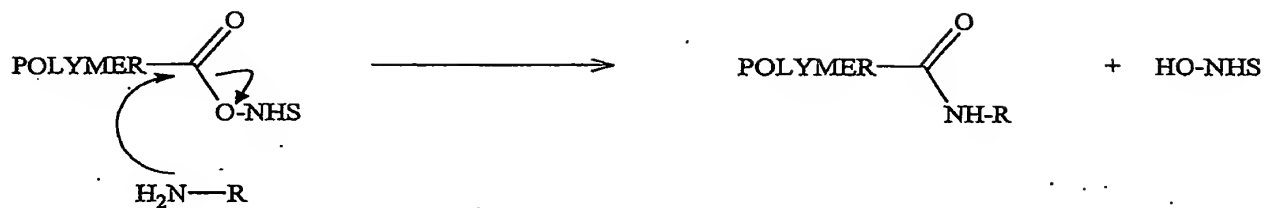


Figure 2

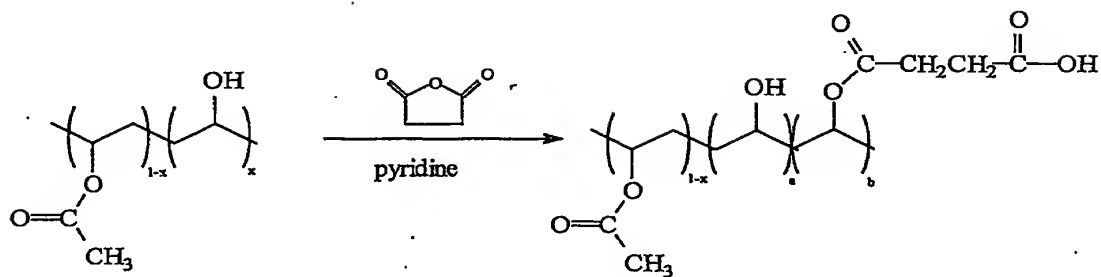


Figure 3

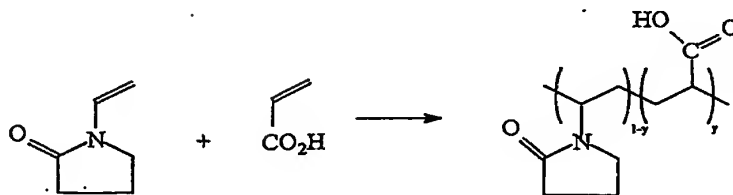


Figure 4

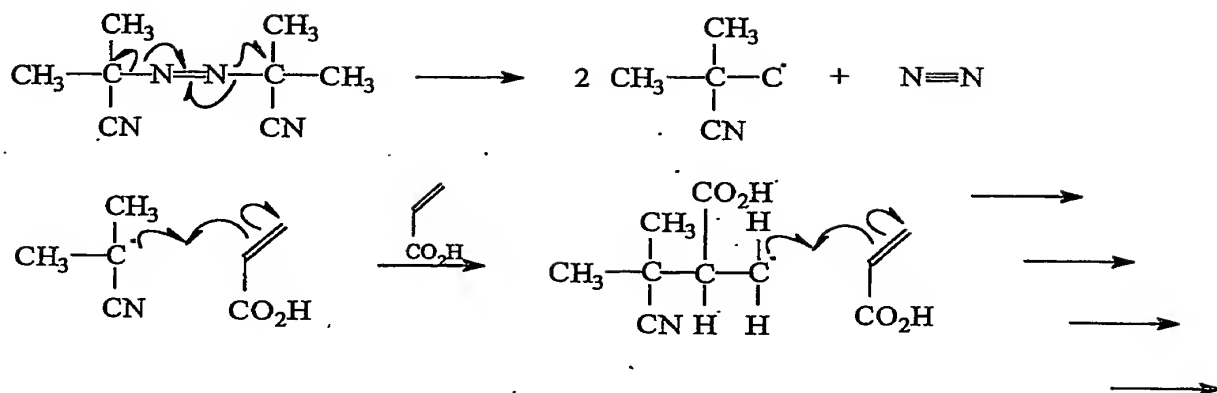


Figure 5

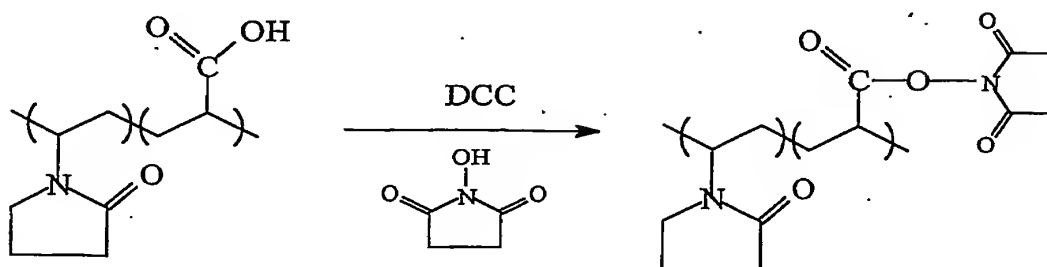
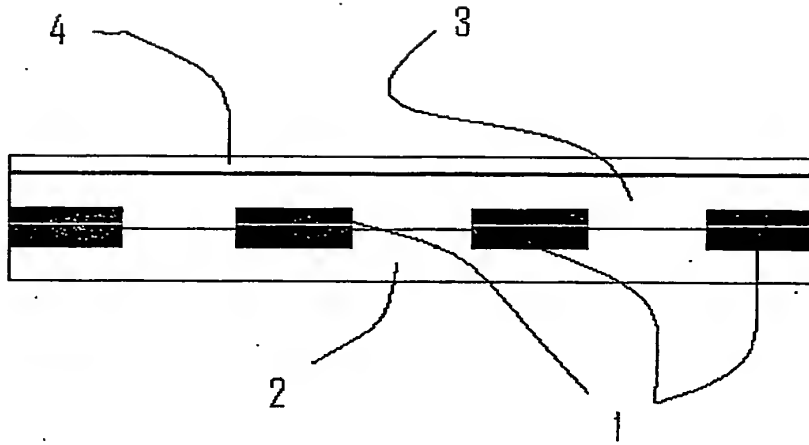


Figure 6





**This Page is Inserted by IFW Indexing and Scanning  
Operations and is not part of the Official Record**

**BEST AVAILABLE IMAGES**

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images include but are not limited to the items checked:

- ☐ BLACK BORDERS
- ☐ IMAGE CUT OFF AT TOP, BOTTOM OR SIDES
- ☐ FADED TEXT OR DRAWING
- ☐ ~~BLURRED OR ILLEGIBLE TEXT OR DRAWING~~
- ☐ SKEWED/SLANTED IMAGES
- ☐ COLOR OR BLACK AND WHITE PHOTOGRAPHS
- ☐ GRAY SCALE DOCUMENTS
- ☐ ~~LINES OR MARKS ON ORIGINAL DOCUMENT~~
- ☐ REFERENCE(S) OR EXHIBIT(S) SUBMITTED ARE POOR QUALITY
- ☐ OTHER: \_\_\_\_\_

**IMAGES ARE BEST AVAILABLE COPY.**

**As rescanning these documents will not correct the image problems checked, please do not report these problems to the IFW Image Problem Mailbox.**